PEPTIDE-ASG4SG3 - MHC & CHAIN REGIONS
Linker

FIG. 1A

- COOH

!

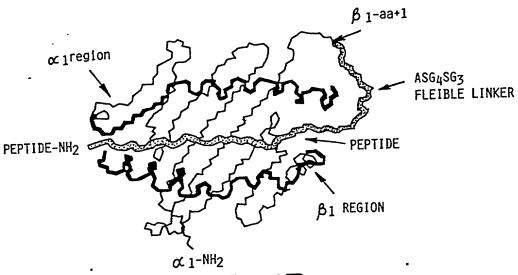
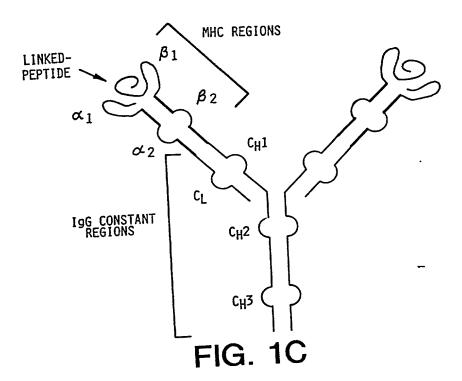
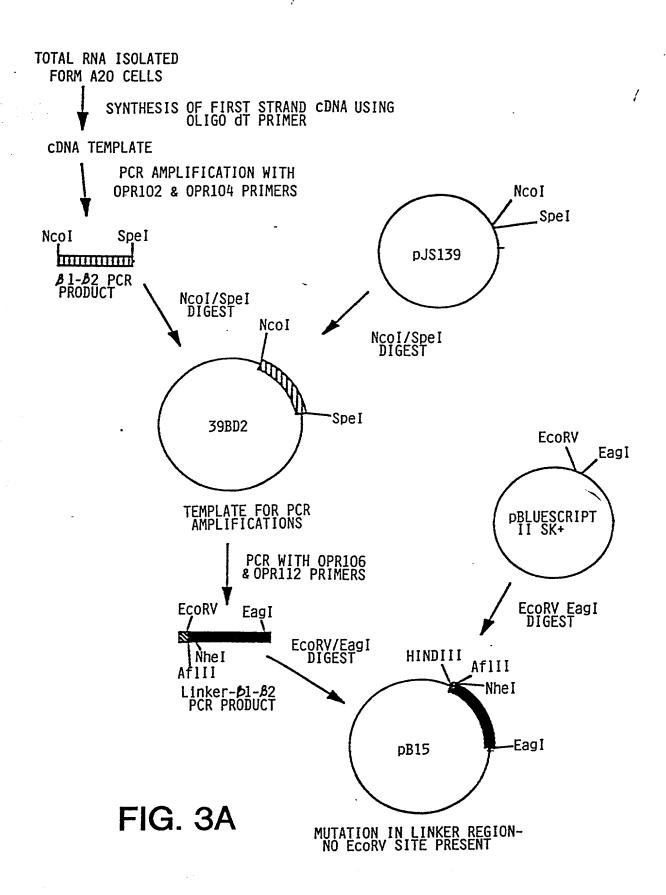
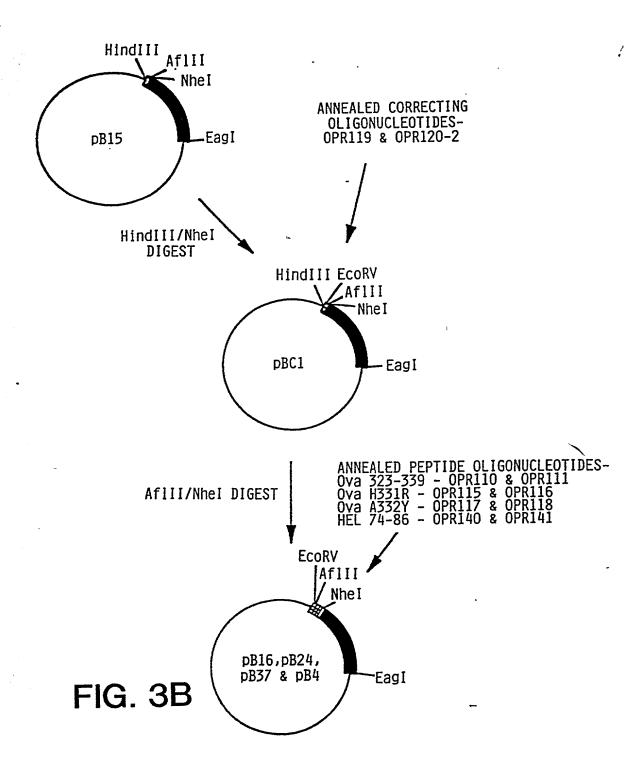


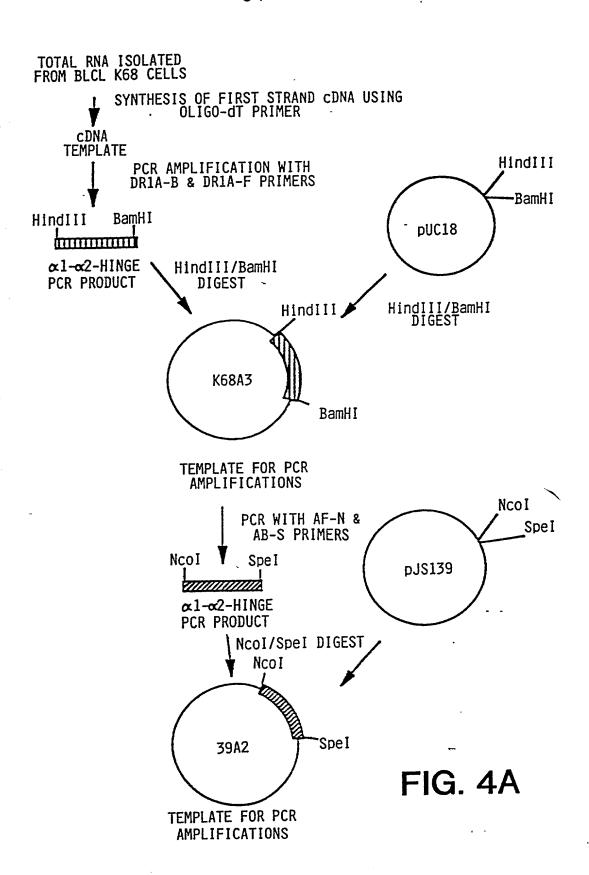
FIG. 1B

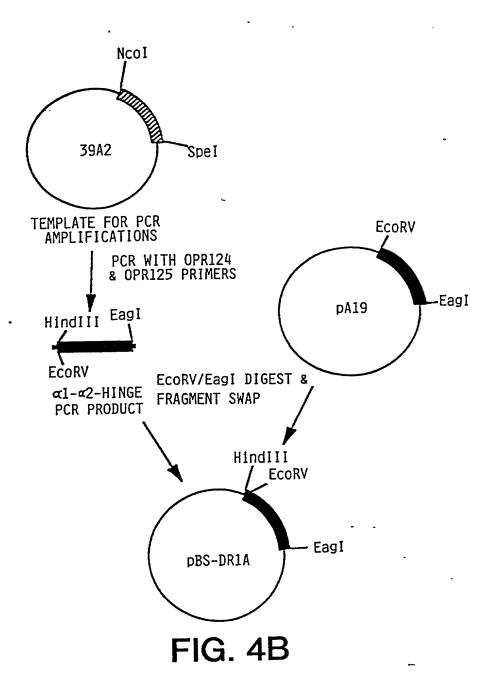


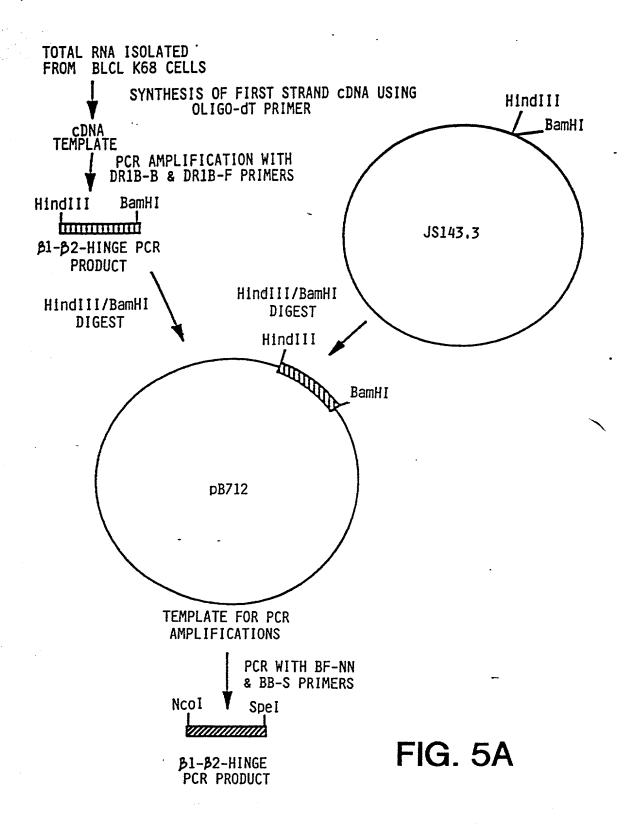
TOTAL RNA ISOLATED FROM A20 CELLS SYNTHESIS OF FIRST STRAND CDNA USING OPRIOI OLIGONUCLEOTIDE PRIMER I-A d **∝**CHAIN SPECIFIC CDNA NcoI PCR AMPLIFICATION WITH OPRIO1 & OPRIO0 PRIMERS SpeI SpeI NcoI pJS139 α1-α2 PCR PRODUCT NcoI/SpeI DIGEST NcoI NcoI/SpeI DIGEST SpeI 39AD2 **EcoRV** EagI TEMPLATE FOR PCR AMPLIFICATIONS PCR WITH OPR107 & OPR108 PRIMERS pBLUESCRIPT II SK+ **EcoRV** EagI EcoRV/EagI DIGEST α1-α2 PCR PRODUCT EcoRV/EagI DIGEST **EcoRV** - Eagl pA19 FIG. 2











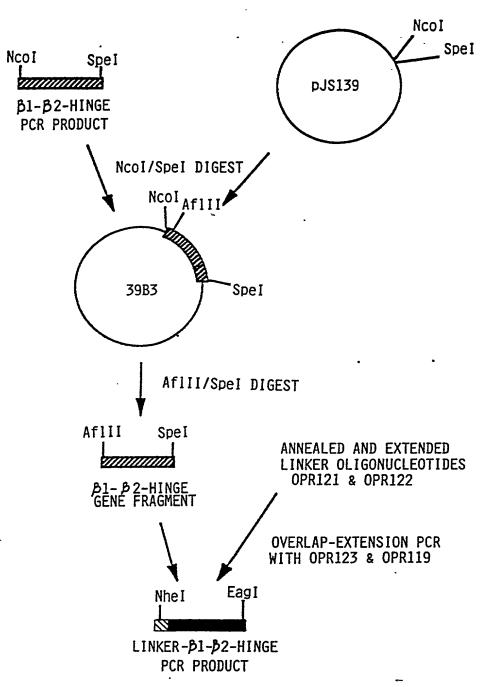
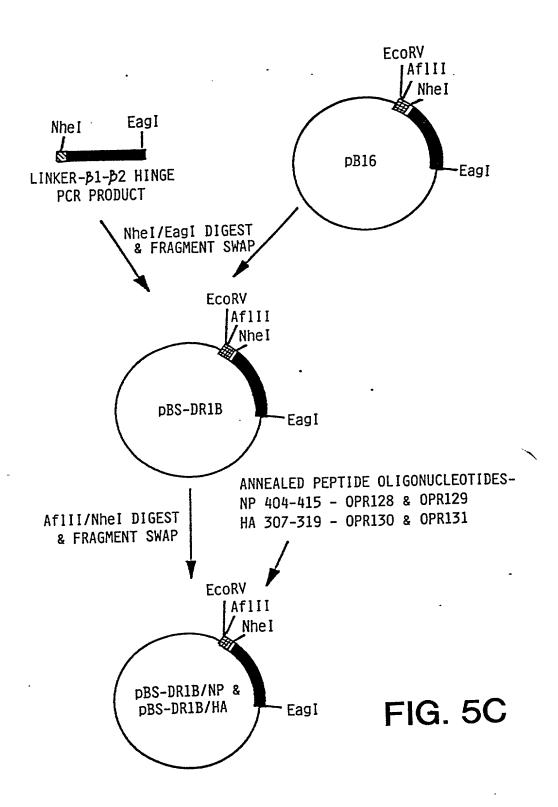
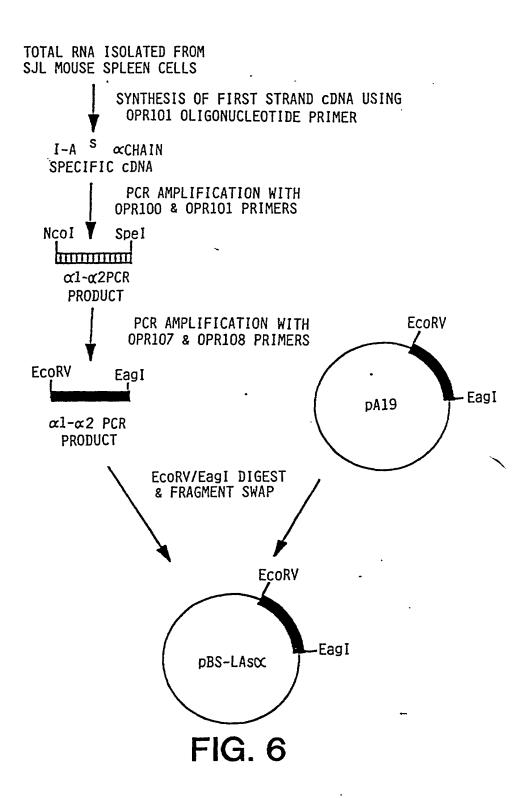
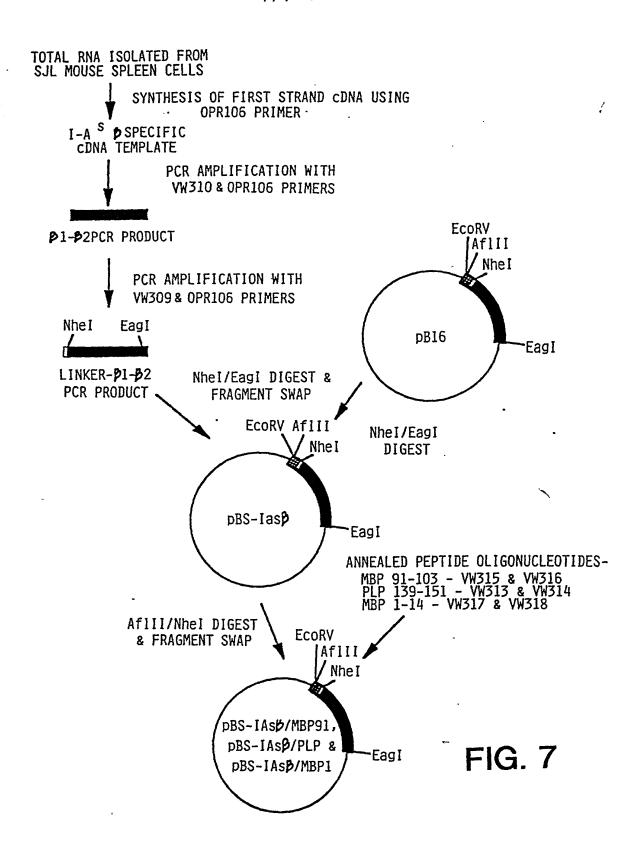


FIG. 5B







12 / 69 I-Ad/I-AS PCR PRIMERS AND CLONING OLIGONUCLEOTIDES (RESTRICTION SITE ARE UNDERLINED). OPR100 5'-GGG GGG GC<u>C ATG G</u>CC GAA GAC GAC GAC ATT GAG GCC GAC-3' OPRIO1 5'-GCG GCG <u>ACT AGT</u> CCA GTG TTT CAG AAC CGG CTC-3' OPR107 5'-CCC CCC GAT ATC TCA GCT TCC AGC AGT GGA GAC GAC ATT GAG GCC G-3' OPR108
5'-CCC CCC CGG CCG CTA CTT ACG TTT CCA GTG TTT CAG AAC CGG
C-3' OPR102 5'-GGG GGG G<u>CC ATG G</u>CC GGA AAC TCC GAA AGG CAT TTC G-3' OPR104 5'-GCG GCG <u>ACT AGT</u> CCA CTC CAC AGT GAT GGG GC-3' OPR106 5'-CCC CCC <u>CGG CCG</u> TAC CTG AGG ACC ACT CCA CAG TGA TGG-3' OPRI12 5'-CCC CCC <u>GAT ATC</u> ACA GGT GTC TTA AGT <u>GCT AGC</u> GGA GGG GGC GGA AGC GGC GGA GGG GGA AAC TCC GAA AGG CAT TTC-3' OPRI19 5'-<u>AGC T</u>TG ATA TCA CAG GTG TCT TAA GTG GA<u>G</u>-3'. OPR120-2 5'-<u>CTA GC</u>T CCA CTT AAG ACA CCT GTG ATA TC<u>A</u>-3' VW310 5'-TCC GGA GGC GGC GGA GAC TCC GAA AGG CAT TTC G-3' VW309 5'-CGA TCG CTA GCG GCG GTG GTG GTG GCG GCG GAG-3' OPR136 5'-CCC CCC <u>AGG CTT CCC GGG</u> CCA CCA TGC CGT GCA GAG CTC TG-3' OPR139 5'-CCC CCC <u>GAG CTC</u> <u>GAA TTC</u> TCA TAA AGG CCC TGG GTG TCT G-3' OPR132
5'-CCC CCC AAG CTT CCC GGG CCA CCA TGG CTC TGC AGA TCC CCA GC-3' OPR133 5'-CCC CCC A<u>CT TAA G</u>GT CCT TGG GCT GCT CAG CAC C-3' OPR134 5'-CCC CCC <u>CCA TCA CTG TGG</u> AGT GGA GGG-3' OPR135 5'-CCC CCC <u>GAG CTC GAA TTC</u> TCA CTG CAG GAG CCC TGC TGG-3'

FIG. 8A

HLA-DRI PCR PRIMERS AND CLONING OLIGONUCLEOTIDES. DRIA-F
5'-GGG GGG AAG CTT ATG ATC AAA GAA GAA CAT GTG ATC ATC-3' DRIA-B
5'-GCG GCG GGA TCC GTT CTC TGT AGT CTC TGG GAG AGG-3' DRIB-F 5'-GGG GGG AAG CTT ATG GGG GAC ACC CGA CCA CGT TTC TTG TGG CAG C-3' AF-N 5'-GGG GGG G<u>CC ATG G</u>CC ATC AAA GAA GAA CAT GTG ATC ATC-3' AB-S 5'-GCG GCG <u>ACT AGT</u> GTT CTC TGT AGT CTC TGG GAG AGG-3' 5'-GGG GGG AAG CTT GAT ATC TCA GCT TCC AGC AGT AGT ATC AAA GAA CAT GTG ATC-3' 5'-GGG GGG CGG. CCG CTA CTT ACG TTT CTC TGG GAG AGG GCT TGG AGC-3' DRIB-B 5'-GCG GCG <u>GGA TCC</u> CTT GCT CTG TGC AGA TTC AGA CC-3' BF-NN 5'-GGG GGG GCC ATG GCC GGA TCC GCT AGC GGG GAC ACC CGA CCA CGT TTC TTG-3' BB-S 5'-GCG GCG ACT AGT CTT GCT CTG TGC AGA TTC AGA CCG-3' OPR121 5'-GTT GTC TTA AGT GGA GCT AGC GGA GGG GGC GGG TCC GGA GGT GGT GGG GAC ACC CG-3' **OPR122** 5'-GAA ATG ACA TTC AAA CTT CAG CTG CCA CAA GAA ACG TGG TCG GGT GTC CCC ACC ACC-3' 5,-GGG GGG CGG CCG TAC CTG AGG ACT TGC TCT GTG CAG ATT CAG-

FIG. 8B

PEPTIDE OLIGONUCLEOTIDES.

Ova 323-339 OPR110 5'-TTA AGT ATC TCT CAG GCT GTT CAC GCT GCT GAA ATC AAC GAA GCT GGT CGT \underline{G} -3'

OPR111
5'-CTA GCA CGA CCA GCT TCG TTG ATT TCA GCC TGA GCA GCG TGA
ACA GCC TGA GAG ATA C-3'

Ova H331R OPR115 $5^\prime\text{-}TTA$ AGT ATC TCT CAG GCT GTT CAC GCT GCT CGG GCT GAA ATC AAC GAA GCT GGT CGT $\underline{G}\text{-}3^\prime$

OPR116 5'-CTA GCA CGA CCA GCT TCG TTG ATT TCA GCC CGA GCA GCG TGA ACA GCC TGA GAG ATA \underline{C} -3'

Ova A332Y OPR117 $5^{\prime}-TTA$ AGT ATC TCT CAG GCT GTT CAC GCT GCT CAC TAC GAA ATC AAC GAA GCT GGT CGT $\underline{G}-3^{\prime}$

OPR116 5'-CTA GCA CGA CCA GCT TCG TTG ATT TCA TAG TGA GCA GCG TGA ACA GCC TGA GAG ATA \underline{C} -3'

HEL 74-86
OPR140
5'-TTA AGT AAC CTG TGC AAC ATC CCC TGC AGC GCC CTG CTG AGC
TCC G-3'

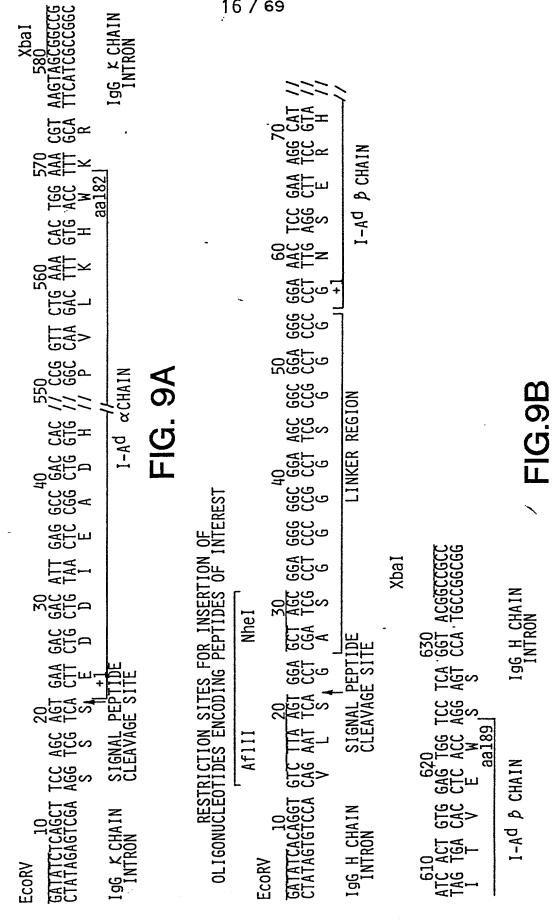
OPR141 5'-CTA GCG GAG CTC AGC AGG GCG CTG CAG GGG ATG TTG CAC AGG TTA \underline{C} -3'

NP 404-415 OPR128 $5'-\overline{TTA}$ AGT CAG ATC AGC GTG CAG CCC GCC TTC AGC GTG CAG \underline{G} -3'

FIG. 8C

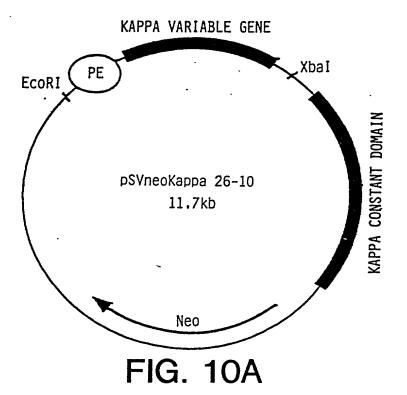
5'-CTĂ GCC TGC ACG CTG AAG GCG GGC TGA ACG CTG ATC TGA C-3' HA 307-319 OPRISO 5'-TTA AGT CCC AAG TAC GTG AAG CAG AAC ACC CTG AAG CTG GCC ACC G-3' 5'-CTA GCG GTG GCC AGC TTC AGG GTG TTC TGC TTC ACG TAC TTG GGA C-3' MBP 91-103 VW315 57-TTA AGT CAC TAT GGC TCC CTG CCG CAG AAG TCC CAG CAC GGG CGC G-3' VW316 5'-CTA GCG CGC CCG TGC TGG GAC TTC TGC GGC AGG GAG CCA TAG TGA \underline{C} -3' PLP 139-151 VW313 5'-TTA CAT CAC TCC CTG GGC AAG TGG CTG GGC CAC CCG GAC AAG VW314 5'-CTA GCG AAC TTG TTC GGG TGG CCC AGC CAC TTG CCC AGG GAG TGA \underline{C} -3' MBP 1-14 VW317 $5'-\overline{1}$ TA AGT ATG GCA TCC CAG AAG CGC CCG TCC CAG CGC TCC AAG TAC CTG G-3'VW316 5'-CTA GCC AGG TAC TTG GAG CGC TGG GAC GGG CGC TTC TGG GAT GCC ATA \underline{C} -3'

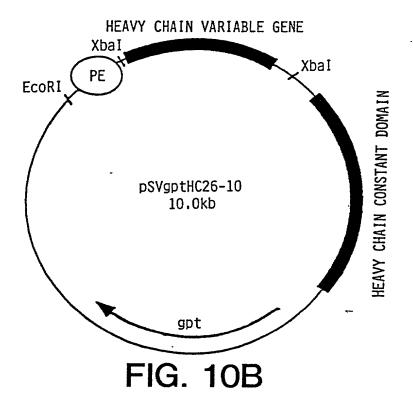
FIG. 8D

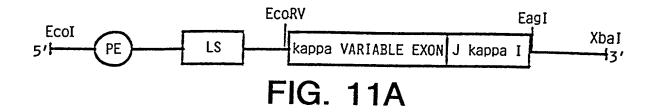


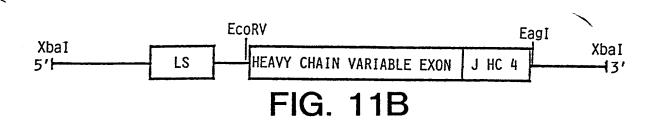
GATATCACAGCT TCC AGC AGT ATC AAA GAA GAA CAT GTG AGT ATC CAT CTT CTT CTT CTT CTT CTT CTT CT	TA CAT H GGG TA CAT TA CAT TA CAT TA CAT TA CGG TA CAT TA		EGG AGG TC AGG GGT CAC AGG TC CCA GGT CCAC TAGG TAGG	CA 6A6 / E CTC 1 IN CCA CCA CCA CCA CCA CCA CCA CCA CCA CC	7 7590 7 767 6 6 6 6 6 6 0 0 0 0 0 0 0 0 0 0 0 0 0 0	ACC CO TTGG GG G	CGT AAGTAG GCA TTCATC I 9G K I 9G K CGT TTC GCA AAG	18 / 69 CCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCC
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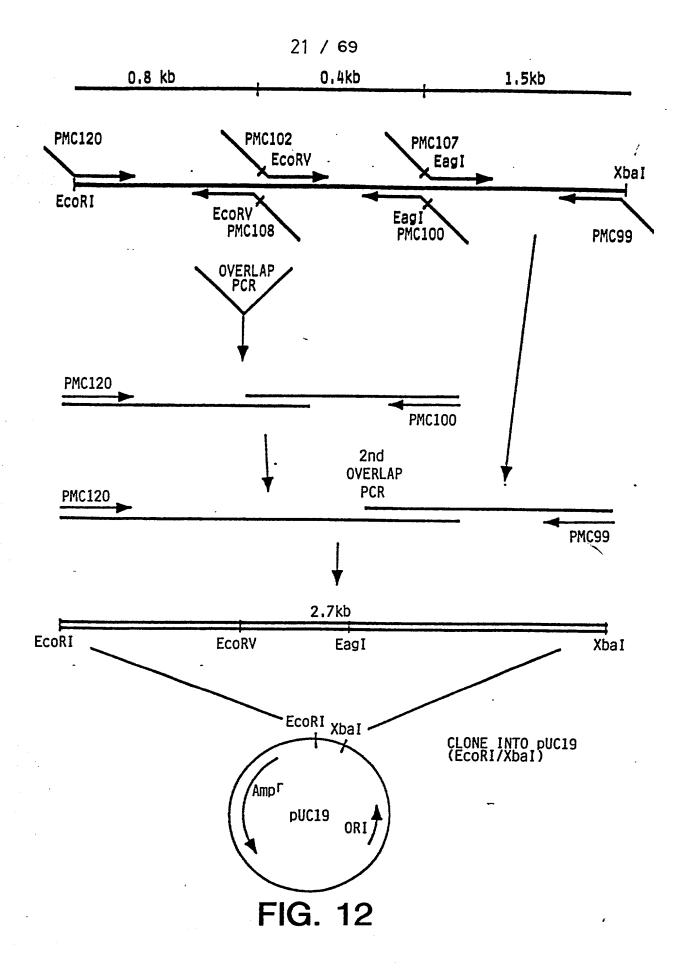
IgG H CHAIN Intron FIG. 9F

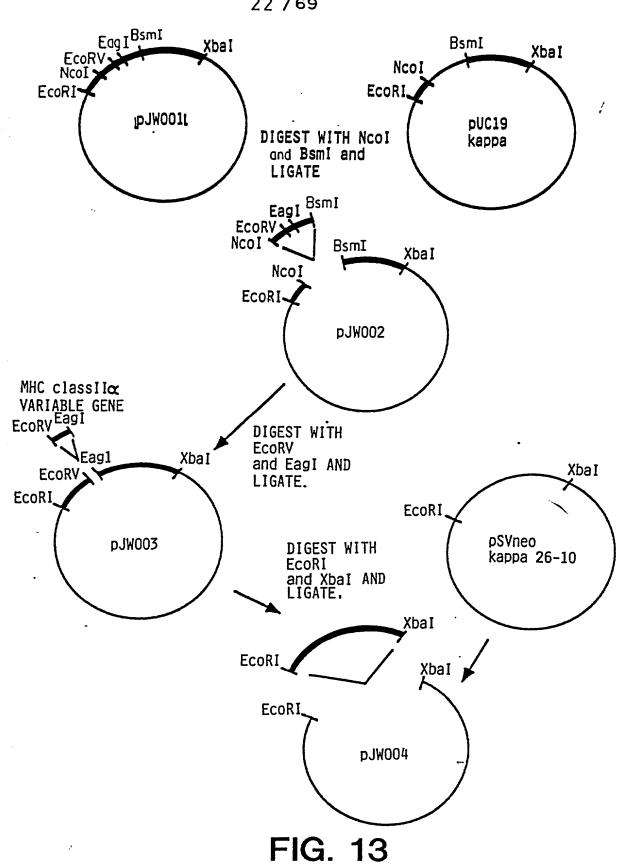


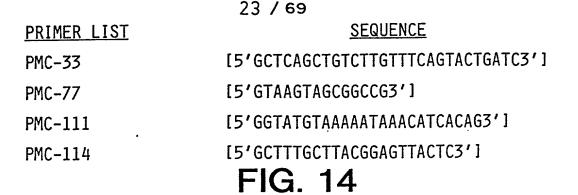


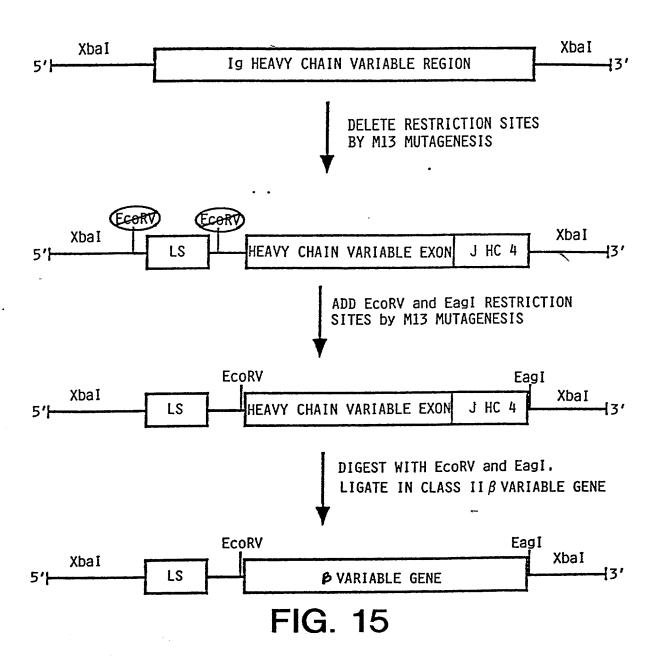


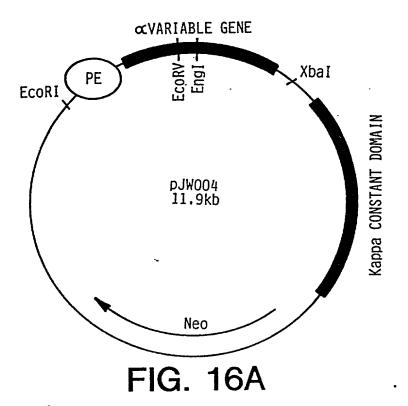


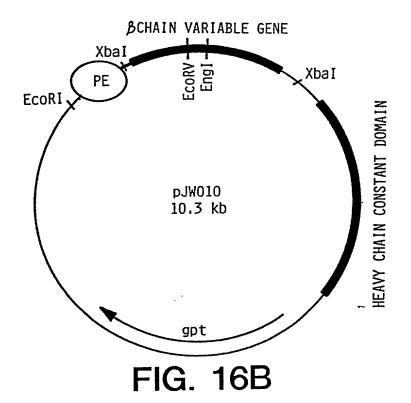


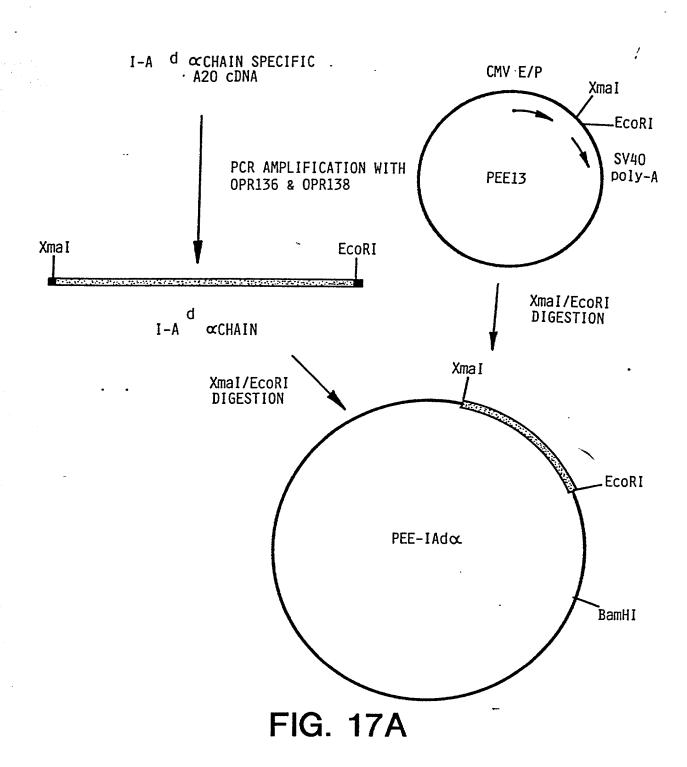


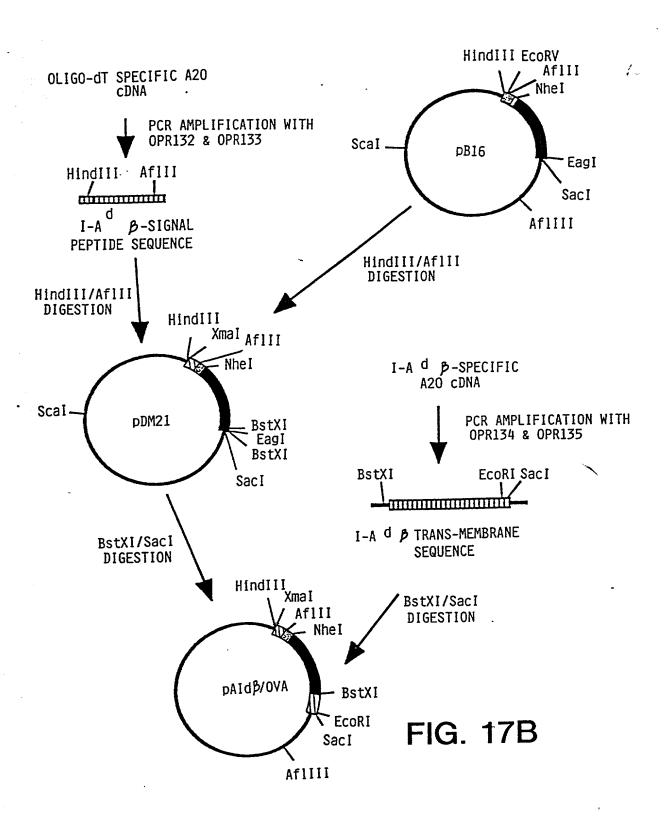


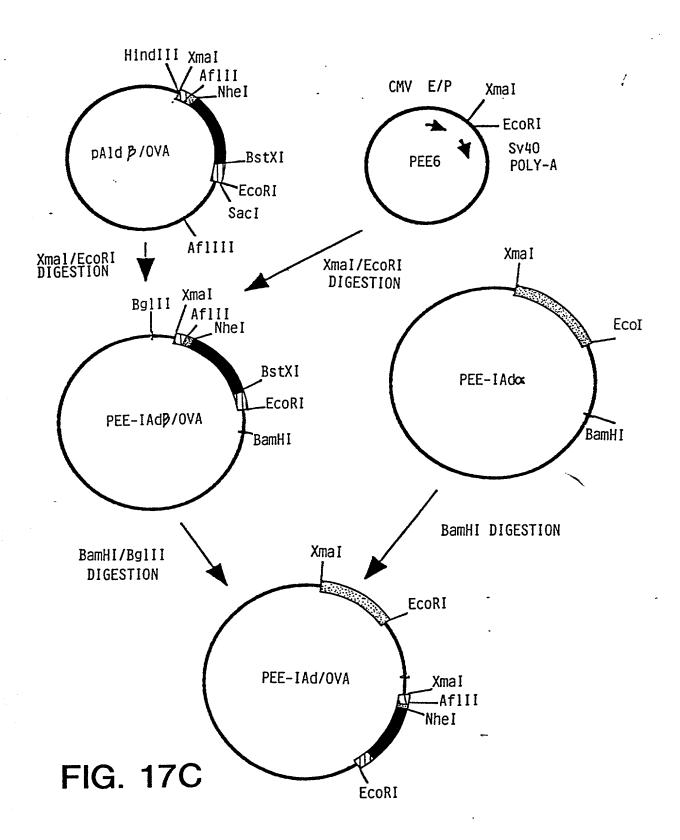


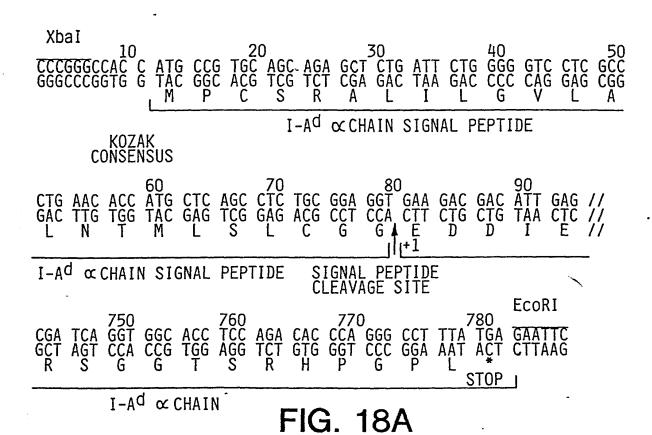












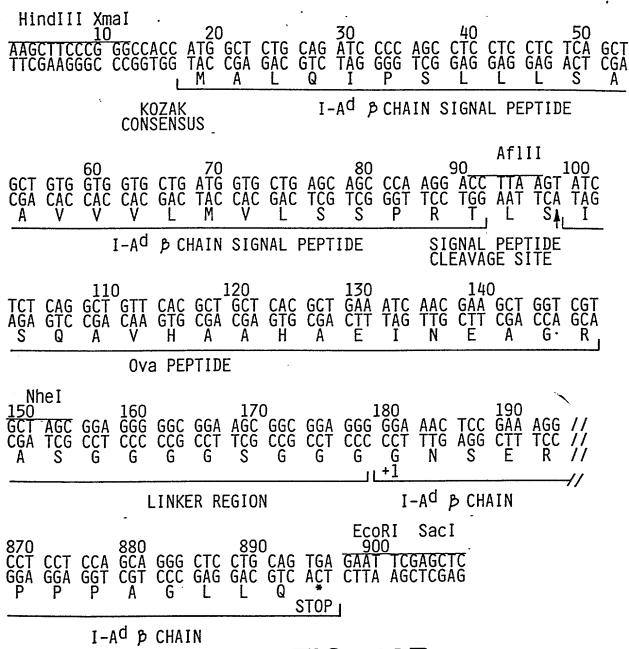
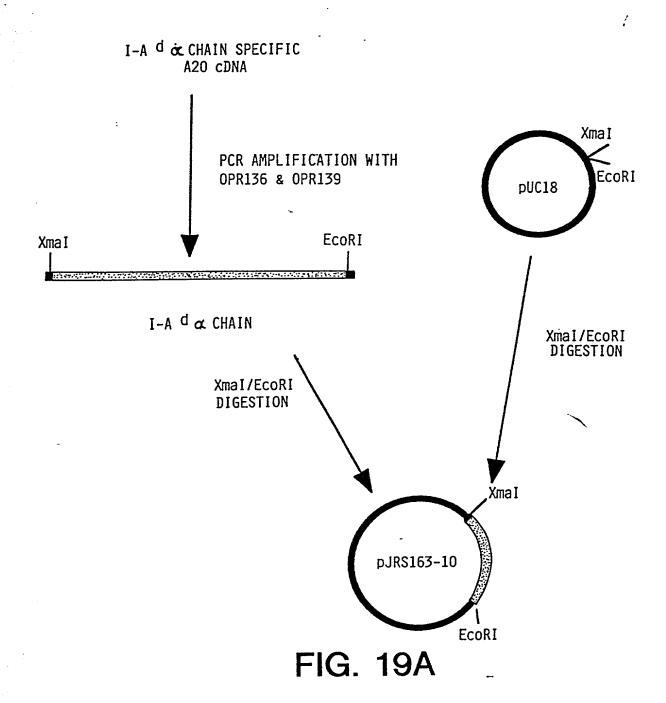
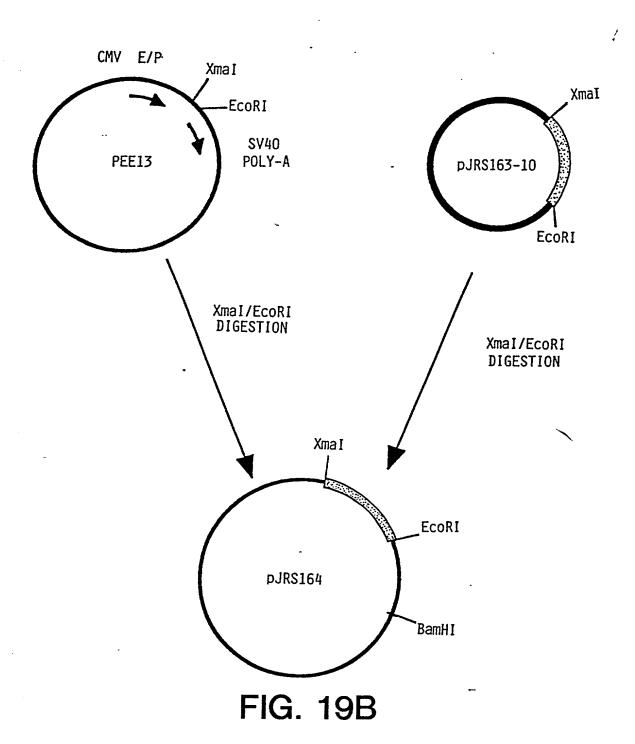
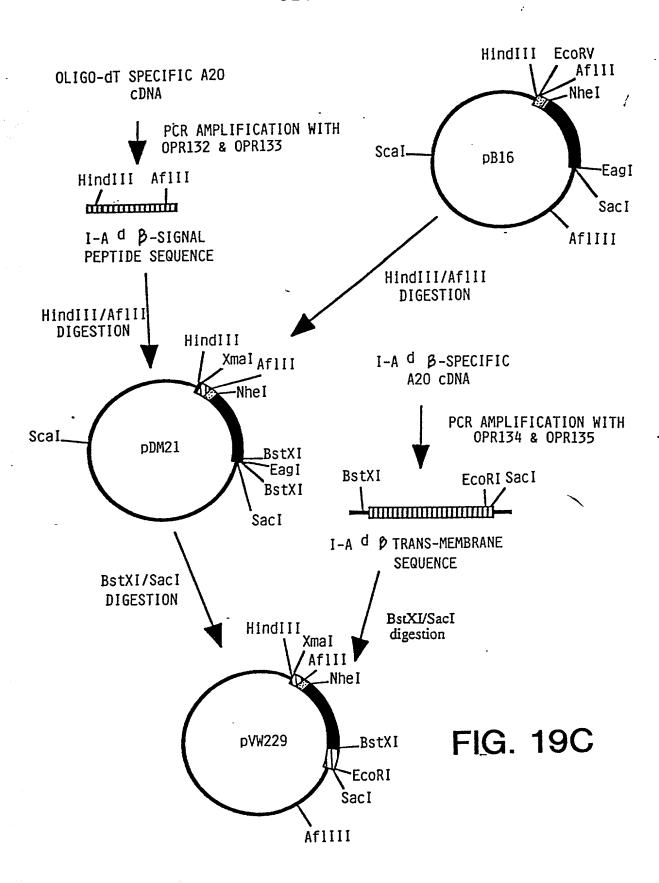
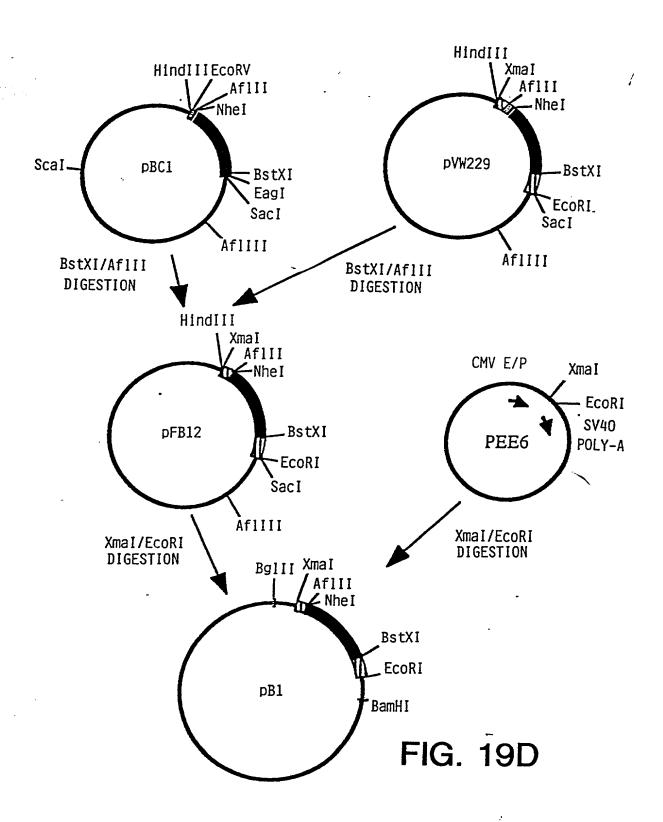


FIG. 18B









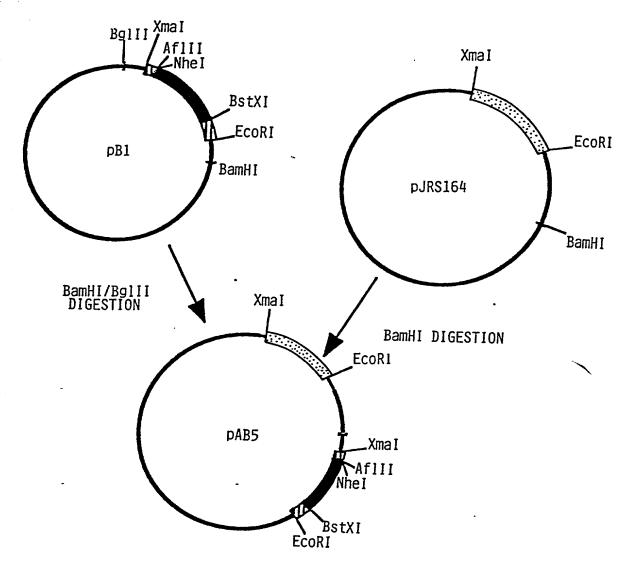
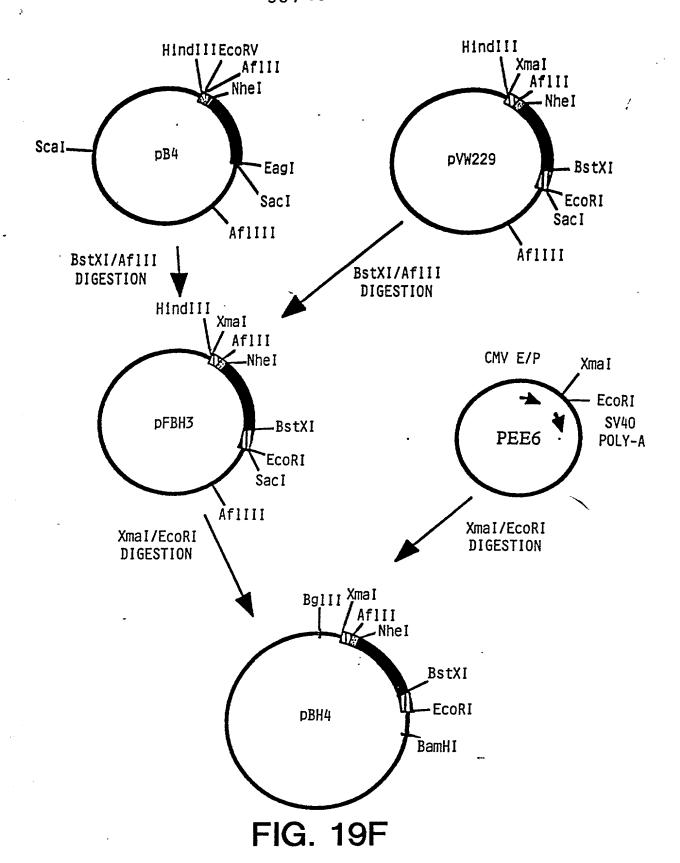


FIG. 19E



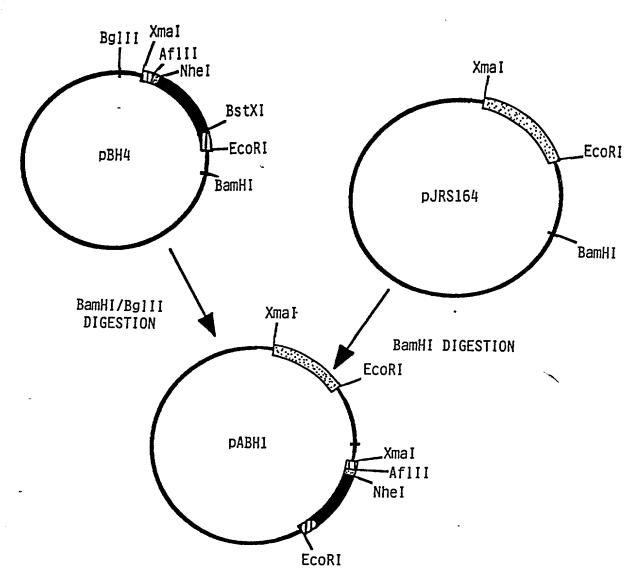


FIG. 19G

OPR132

I-A^d β signal peptide front primer with Kozak consensus for CellTech vector - HindIII/XmaI sites
5'-CCC CCC AAG CTT CCC GGG CCA CCA TGG CTC TGC AGA TCC CCA GC-3'

OPR133

I-A^d β signal peptide back primer with Kozak consensus for CellTech vector - AfIII site 5'-CCC CCC ACT TAA GGT CCT TGG GCT GCT CAG CAC C-3'

OPR134

I-A^d β transmembrane front primer for CellTech vector - BstXI sites 5'-CCC CCC CCA TCA CTG TGG AGT GGA GGG-3'

OPR135

I-A^d β transmembrane back primer for CellTech vector - SstI, EcoRI sites
5'-CCC CCC GAG CTC GAA TCC TCA CTG CAG GAG CCC TGC TGG-3'

OPR136

I-A^d α signal peptide front primer with Kozak consensus for CellTech vector - HindIII/XmaI sites
5'-CCC CCC AAG CTT CCC GGG CCA CCA TGC CGT GCA GAG CTC TG-3'

OPR139

I-A^d α transmembrane primer for CellTech vector - SstI/EcoRI sites 5'-CCC CCC GAG CTC GAA TCC TCA TAA AGG CCC TGG GTG TCT G-3'

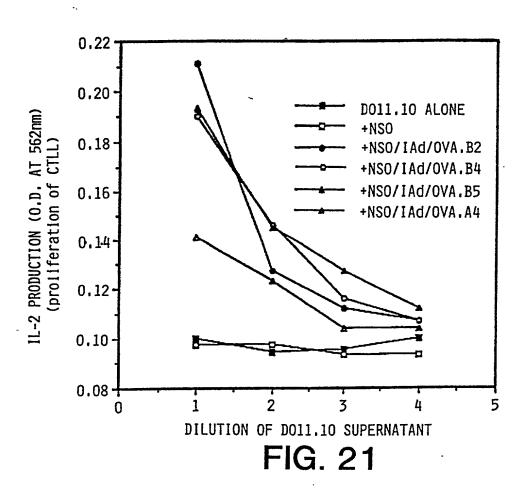
B7-1-2F

Murine B7-1 front primer with Kozak consensus for CloneTech vector - NotI site 5'-CCC CCC CCG CGG CCG CCC CAC CAT GGG ACT GAG TAA CAT TCT C-3'

B7-1-2B

Murine B7-1 BACK primer for CloneTech vector - NotI site 5'-CCC CCC GCG GCC GCT TTA AAA ACA TGT ATC ACT TTT-3'

FIG. 20



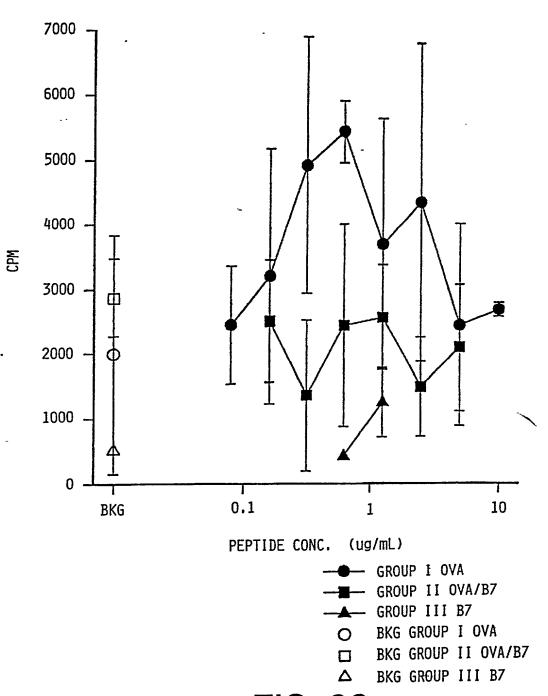
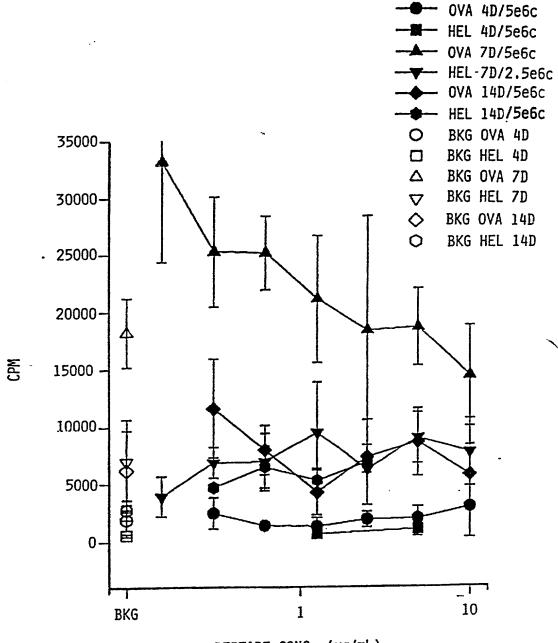


FIG. 22



PEPTIDE CONC. (ug/mL)

FIG. 23

LINKER SEQUENCE LINKED TO PRESENTING PEPTIDE

PEPTIDE BINDING GROOVE



α2 DOMAIN

B2 DOMAIN

SINGLE CHAIN LINKER SEQUENCE

FIG.24

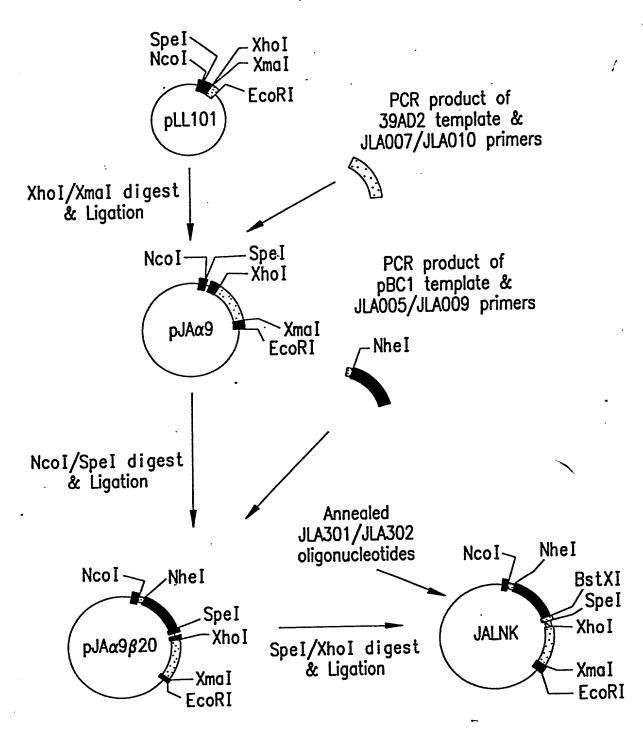
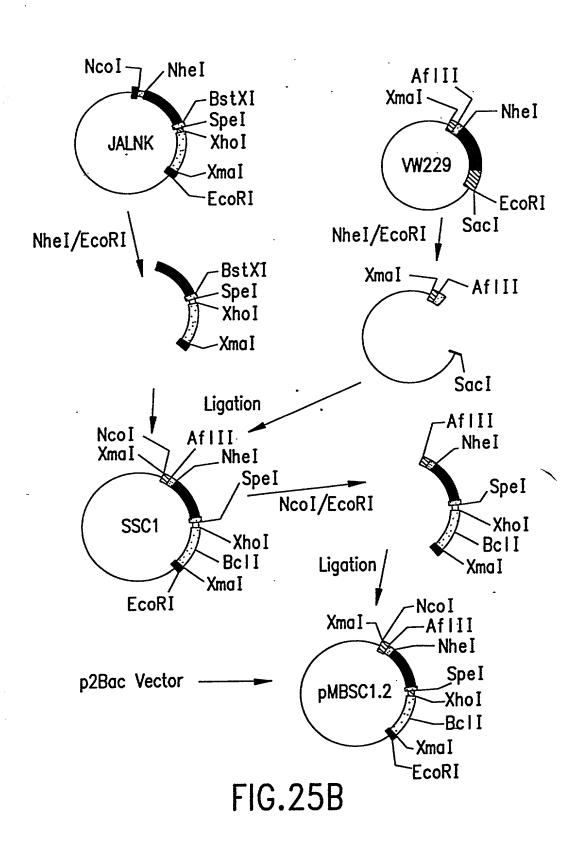


FIG.25A



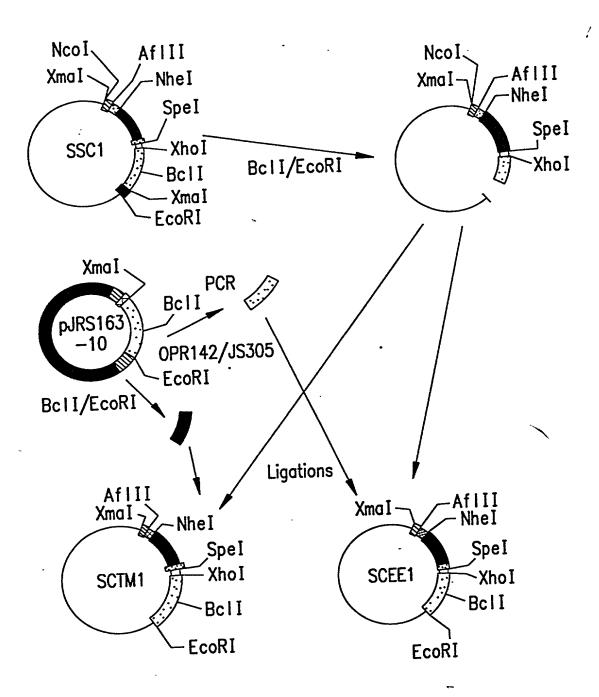


FIG.25C

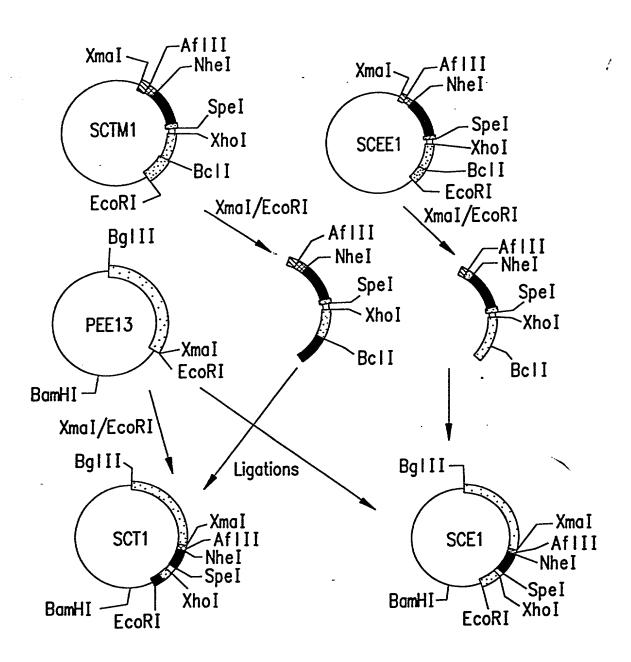


FIG.25D

JLA-005 5'-CCCCCGCCATGGCCGCTAGCGGAGGGGGGGGAAGC-3'

JLA-007 5'-CCCGGGGCCTCGAGTGAAGACGACATTGAGGCCGAC-3'

JLA-009 5'-CCCCCACTAGTCCACTCCACAGTGATGGGGCT-3'

JLA-010 5'-CCCCCCCGGGACCAGTGTTTCAGAACCGGCTCCTC-3'

JLA-301 5'-TCGAGGAACCGCCACCGCCAGAACCGCCGCCACCGGA-ACCACCACCGCCGCTGCCACCGCCACCA-3'

JLA-302 5'-CTAGTGGTGGCGGTGGCAGCGGCGGTGGTTCCGG-TGGCGGCGGTTCTGGCGGTGGCGGTTCC-3'

OPR-142 5'-CTTGGGAATCTTGACTAAGAGG-3'

JS-305 5'-CAGGTCGAATTCTCATTCCATCGGCATGTACTCTTCTT-CCTCCCAGTGTTTCAGAACCGG-3'

FIG.26

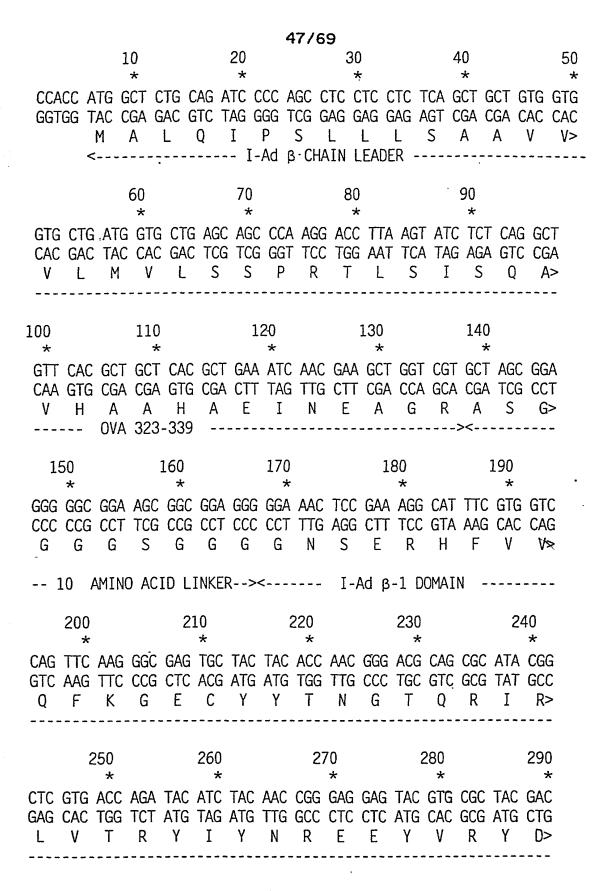


FIG.27A

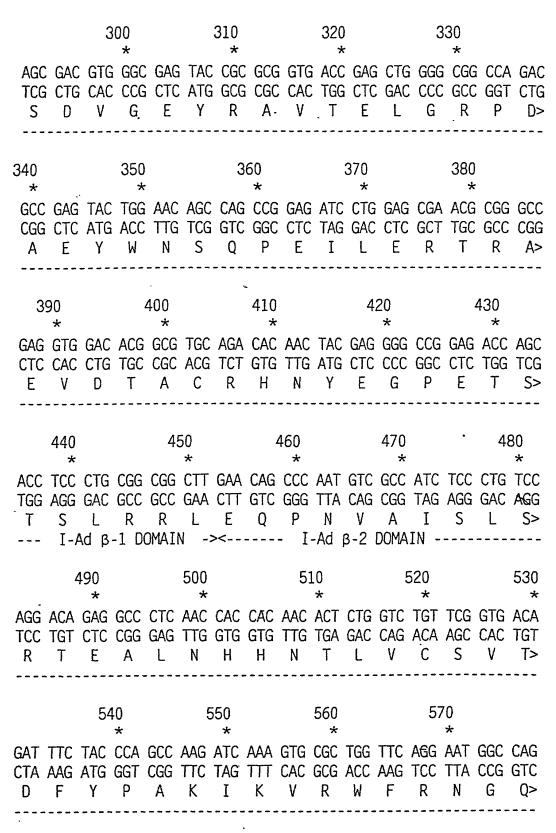


FIG. 27B

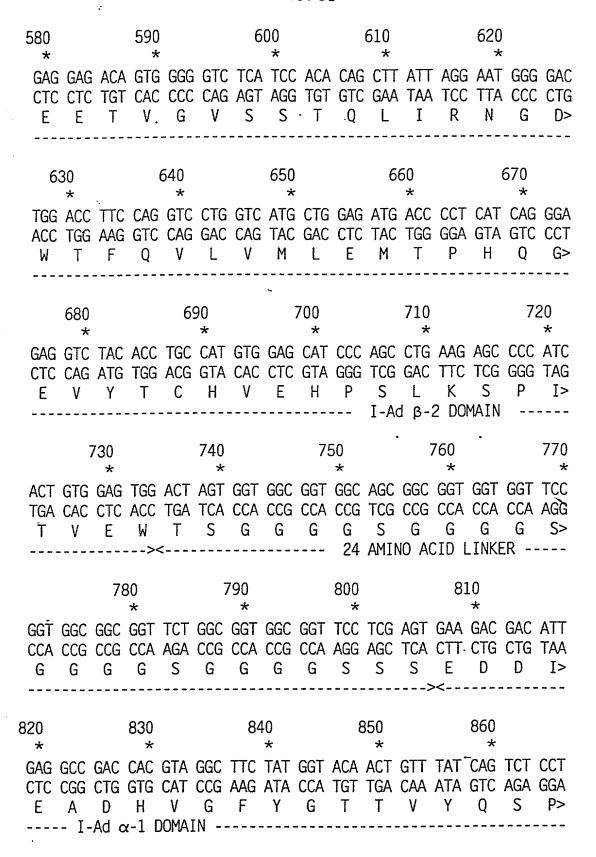


FIG.27C

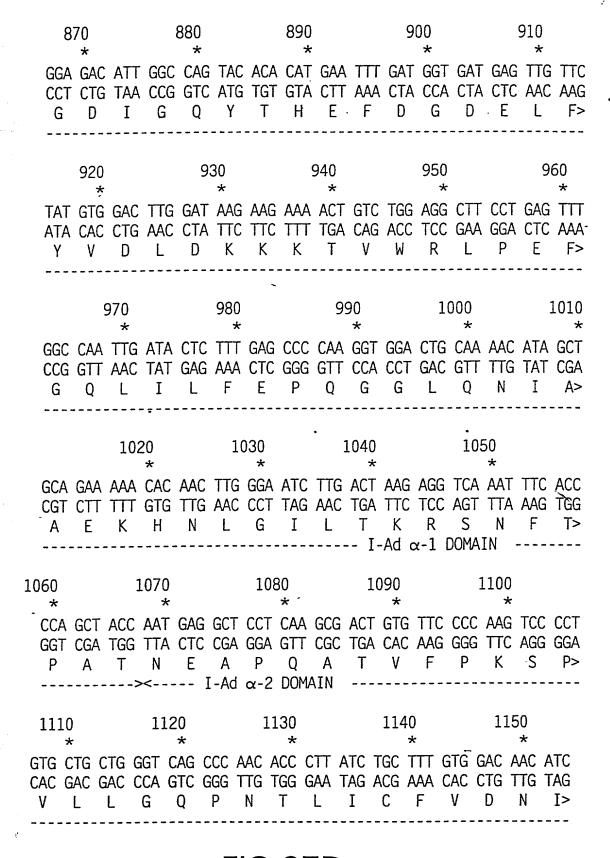


FIG.27D

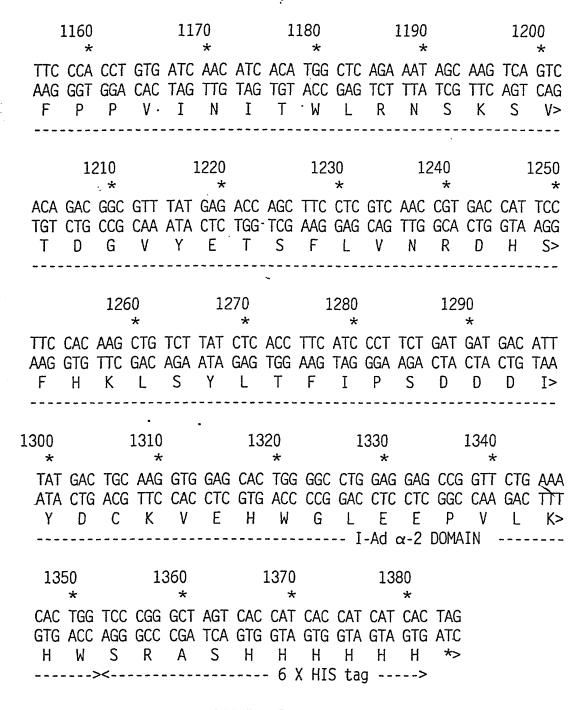


FIG.27E

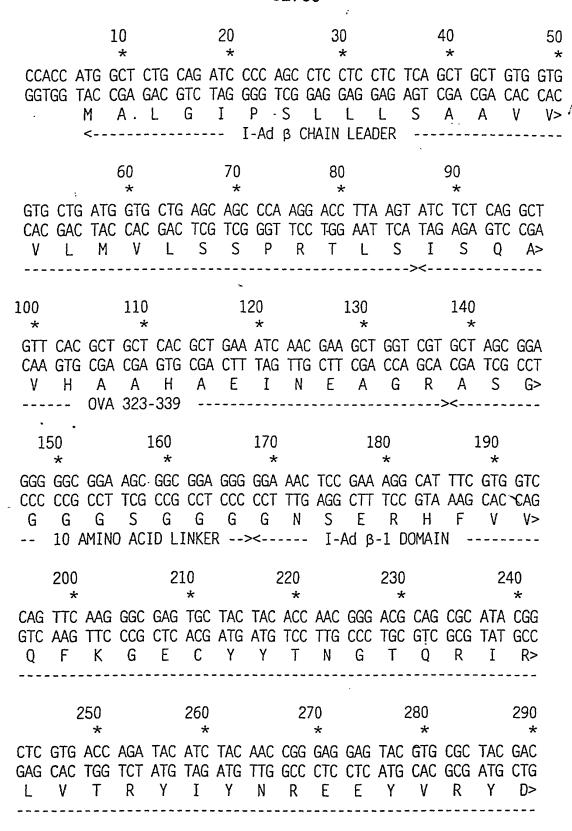


FIG.28A

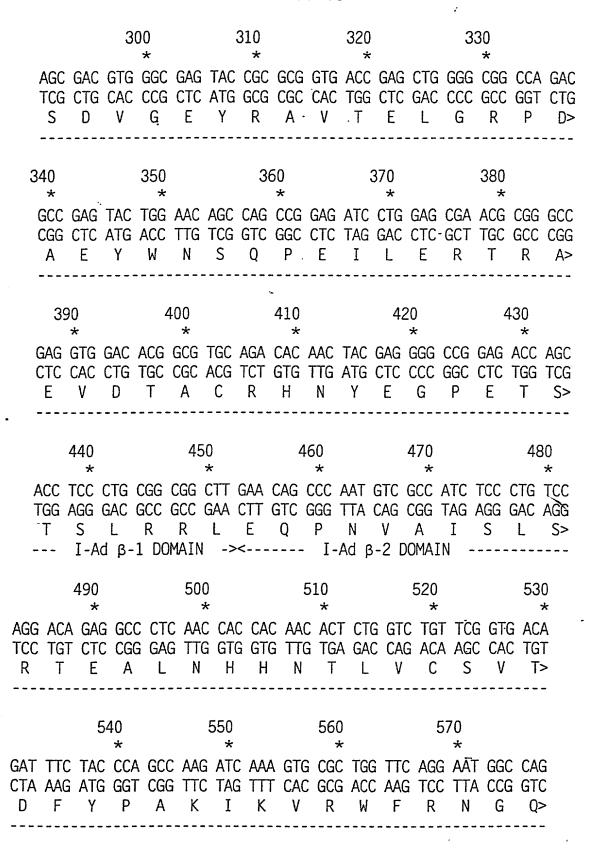


FIG.28B

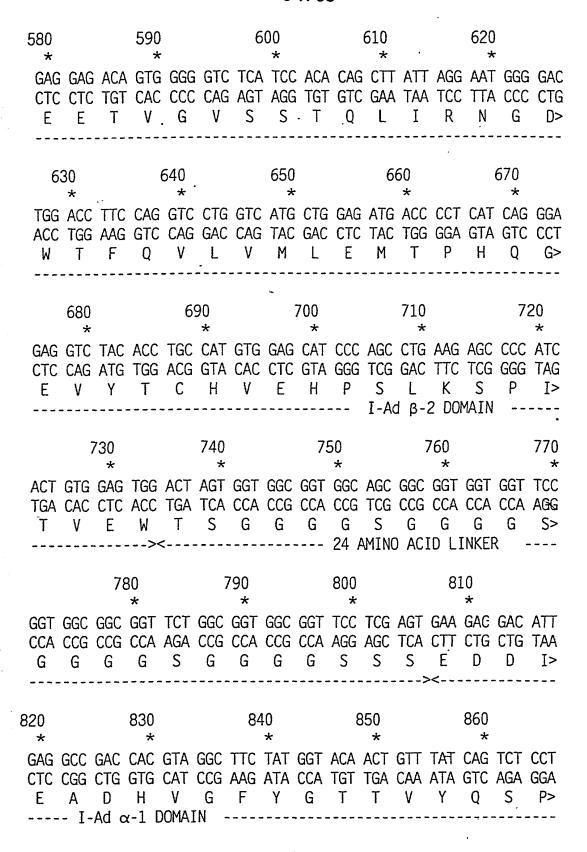


FIG.28C

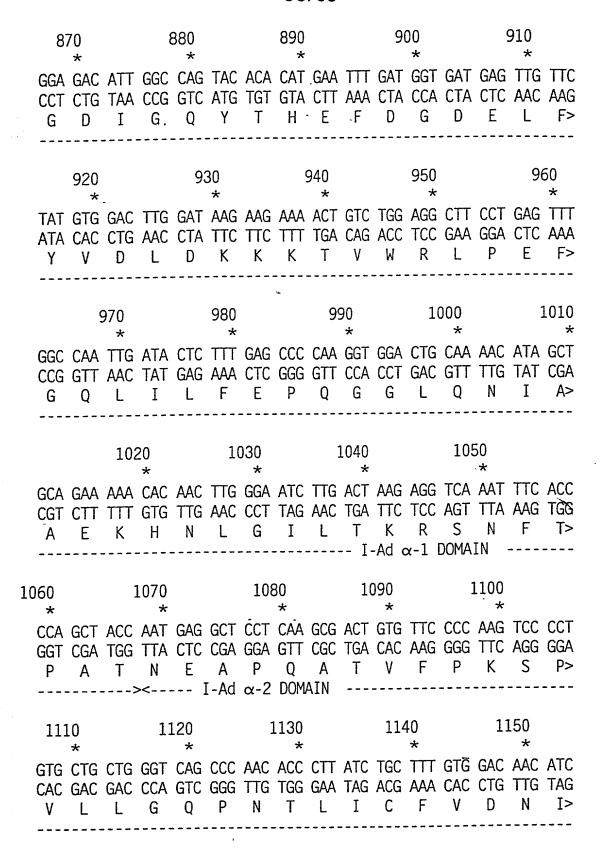


FIG.28D

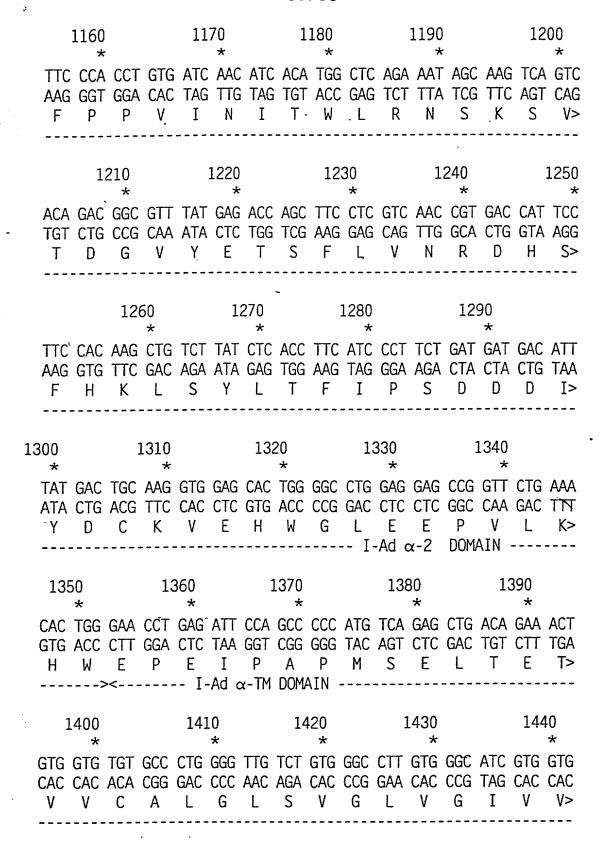


FIG.28E

CAC CCA GGG CCT TTA TGA GTG GGT CCC GGA AAT ACT H P G P L *> - I-Ad α -TM DOMAIN ->

FIG.28F

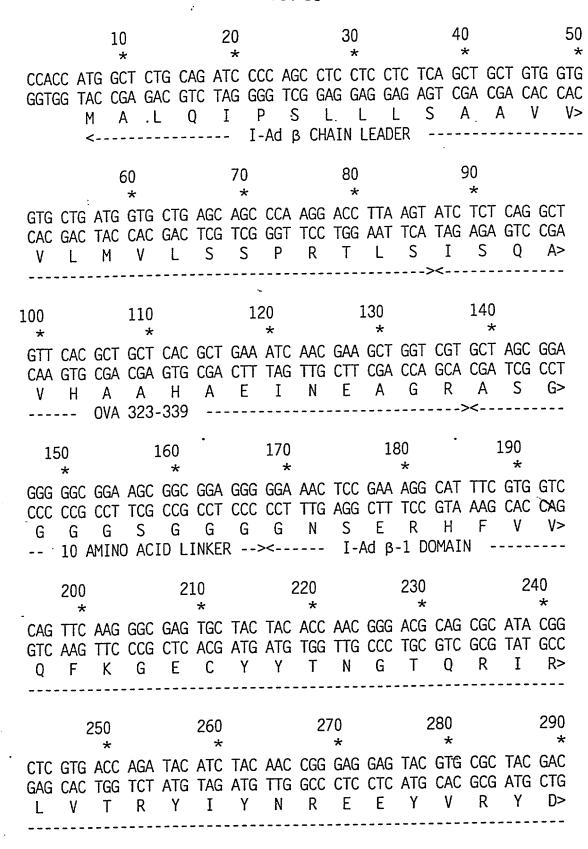


FIG.29A

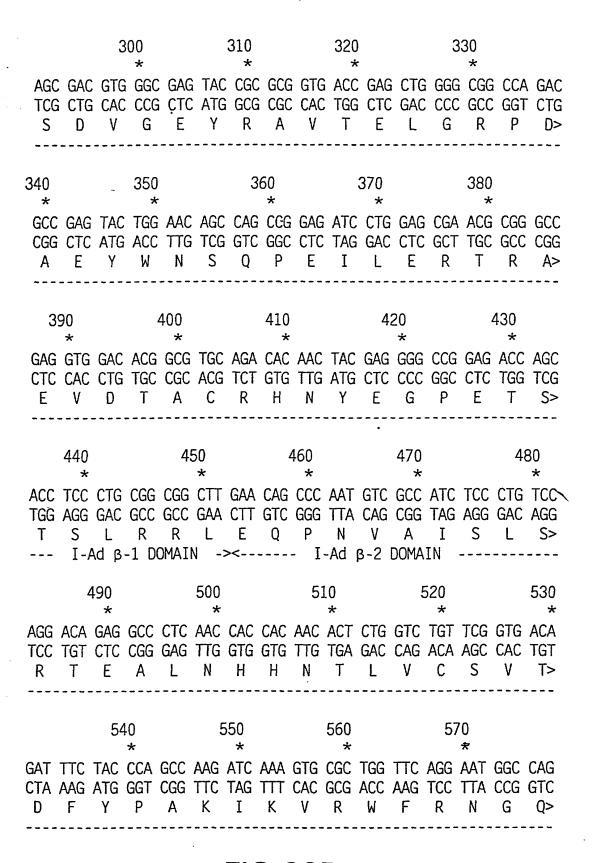


FIG.29B

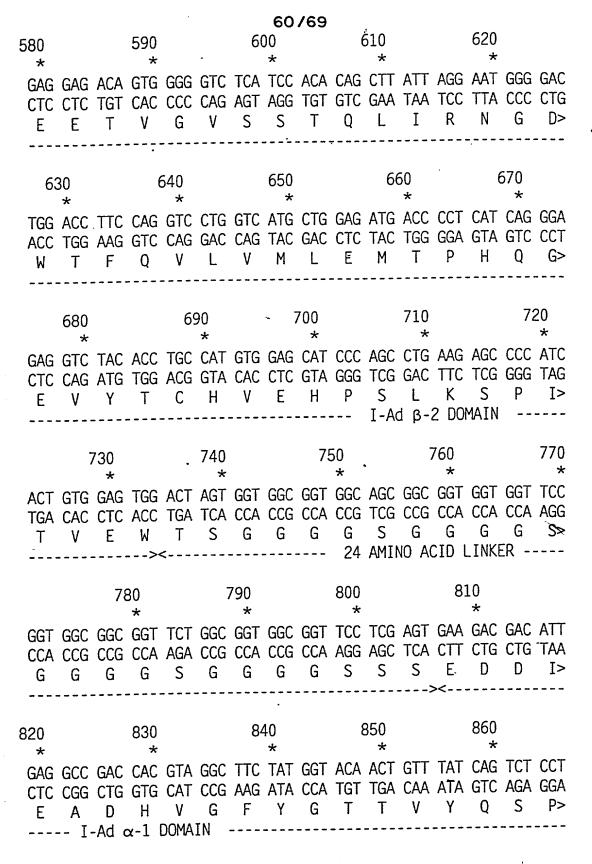


FIG.29C

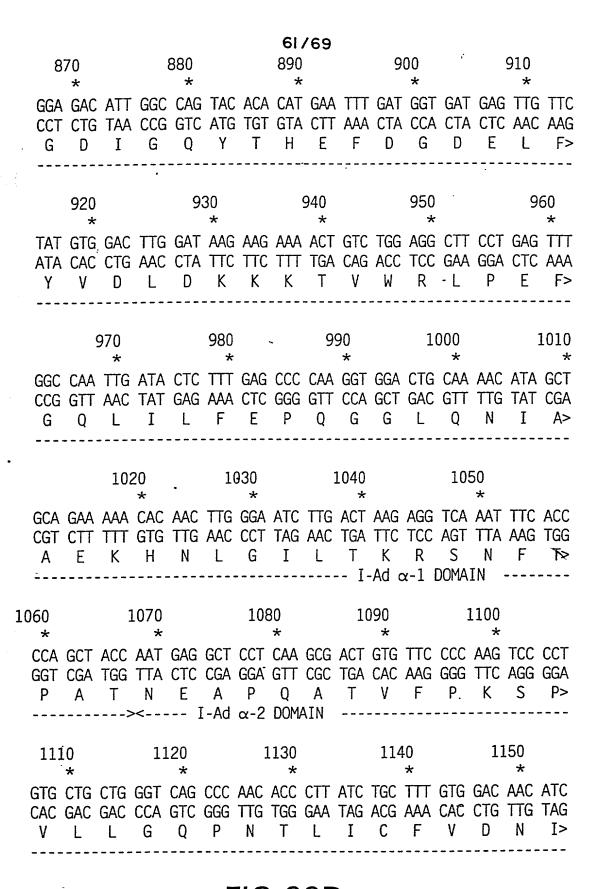


FIG.29D

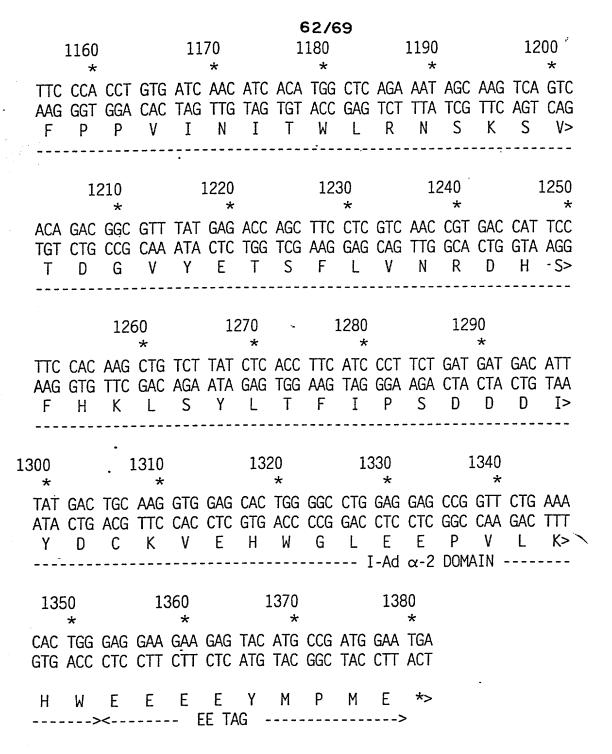
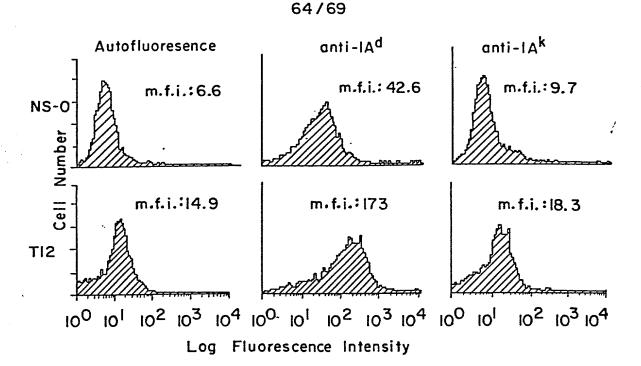


FIG.29E

CCACC ATG GCT CTG CAG ATC CCC AGC CTC CTC TCA GCT GTG GTG GTG CTG ATG GTG CTG AGC CCA AGG ACC TTA AGT ATC TCT CAG GCT GTT CAC	Single chain linker means and a single chain linker means a single chain linke
CTG,	o
GTG V V V V V V V V V V V V V V V V V V	တ
a GGT GGT GGT GGT GGT GGT GGT GGT GGT GG	O
T GTG V gnal p GG G G 1. — 1	0
CT GCA (GAA)	G
S Ad Bo	S G chain
CTC T L L L A J L GITTO	G S Single
ST CG	o
C CTC	g
SC AG	5
AAC N N N N N N N N N N N N N N N N N N	တ
CAGA AATC I Peptide	<u>ق</u>
CTG	5
CCACC ATG GCT CTG CAG ATC CCC AGC CTC CTC CTC TC M A L Q I P S L L L S Kozek	5
C ATC M Edge C TC C	n
CCACC AT M Kozak consensus GCT GCT (A A —	

F16.30



F I G. 31A

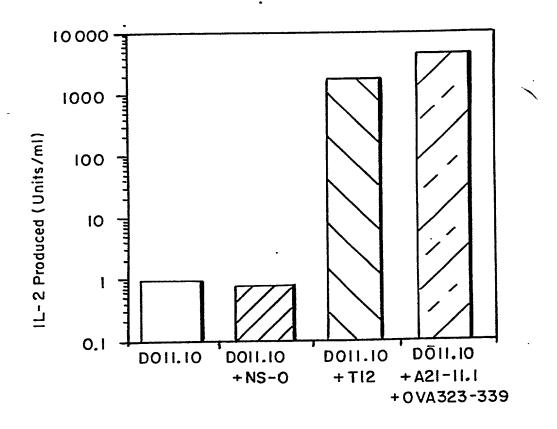


FIG. 31B

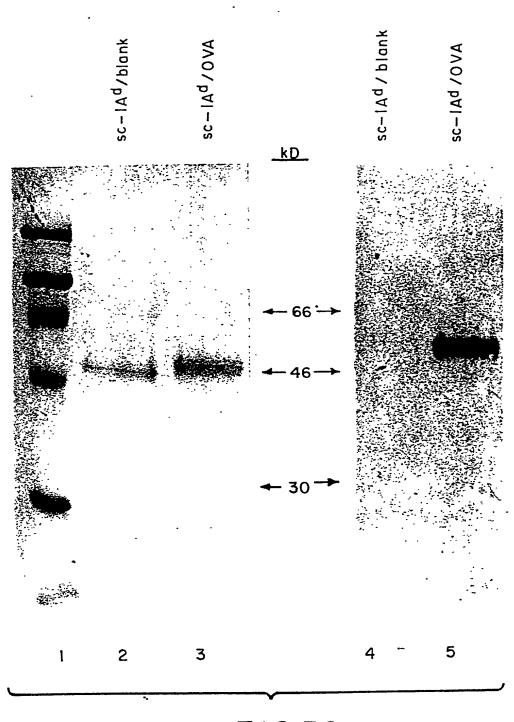


FIG.32

